Application No. 10/518,749 Amendment Dated September 10, 2010

Response Under 37 C.F.R. § 1.116

AMENDMENTS TO THE CLAIMS

 (Currently Amended) A method for producing isolated neural cells, wherein the method comprises:

earrying outculturing primate embryonic stem cells in suspension a suspension culture of primate embryonic-stem cells for a period of 7 to 15 days, in the presence of an astrocyte conditioned medium or ingredients equivalent to the conditioned medium to directly-produce said isolated neural cells.

2. (Cancelled)

- 3. (Previously Presented) The method for producing neural cells according to claim 1. wherein the primate embryonic stem cells are selected from the group consisting of cynomolgus monkey embryonic stem cells and human embryonic stem cells.
- 4. (Currently Amended) The-A method for producing isolated neural cells-according to elaim-1, wherein the method comprises the step of:
- (A) earrying out the suspension culture of culturing primate embryonic stem cells in suspension in the presence of the an astrocyte conditioned medium or ingredients equivalent to the conditioned medium, thereby forming a stem cell sphere (SCS) comprising neural stem cells.
- 5. (Currently Amended) The method for producing neural cells according to claim 4, further comprising carrying out after the step (A), the step of:
- (B) culturing-thesaid stem cell sphere (SCS) obtained in the step (A) in the presence of basic fibroblast growth factor (bFGF) and/or epidermal growth factor (EGF) and in the presence of a cell adhesion molecule, thereby obtaining neural stem cells as cells migrated from SCS
- 6. (Currently Amended) The method for producing neural cells according to claim 5, wherein the culture in the step (B) is carried out in the state of adhesion of the stem cell sphere

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(SCS) obtained in the step (A) is adhered to an adhesive a culture substratum carrying a cell adhesion molecule.

- 7. (Currently Amended) The method for producing neural cells according to claim 1, comprising ewherein said arrying out the step of:
- (A') earrying out the suspension culture of culturing primate embryonic stem cells in suspension is carried out in the presence of the astrocyte conditioned medium-or-ingredients equivalent to the conditioned medium, and in the presence of basic fibroblast growth factor (bFGF) and/or epidermal growth factor (EGF), thereby obtaining neural stem cells in a stem cell sphere (SCS).
- 8. (Currently Amended) The method for producing neural cells according to claim 4, further comprising carrying out after the step (A), the step of:
- culturing the said stem cell sphere (SCS) obtained in the step (A) in the state of adhesion of SCS to an adhesive culture substratum earrying comprising a cell adhesion molecule in the absence of basic fibroblast growth factor (bFGF) and/or epidermal growth factor (EGF) and in the presence of an astrocyte conditioned medium or ingredients equivalent to the conditioned medium, thereby obtaining a neuron.
- 9. (Currently Amended) The method for producing neural cells according to claim 4. further comprising carrying out after the step (A), the steps of:
- (B) culturing the-said stem cell sphere (SCS) obtained in the step (A) in the presence of basic fibroblast growth factor (bFGF) and/or epidermal growth factor (EGF) and in the presence of a cell adhesion molecule; and subsequently
- (C) culturing the SCS obtained in the step (B) in the state of adhesion of SCS to an adhesive culture substratum earrying comprising a cell adhesion molecule and in the absence of bFGF and/or EGF, thereby obtaining glial cells as cells migrated from SCS.
- 10. (Previously Presented) A method for producing a neuron, comprising the step of culturing the neural stem cells obtained by the method according to claim 1 in a state of adhesion

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of the neural stem cell to an adhesive culture substratum carrying a cell adhesion molecule in the absence of basic fibroblast growth factor (bFGF) and/or epidermal growth factor (EGF), and in the presence of the astrocyte conditioned medium or ingredients equivalent to the conditioned medium

- (Currently Amended) Isolated neural stem cells, which are differentiated from an embryonic stem cellsprepared by the method according to claim 1.
- (Currently Amended) The neural stem cells according to claim 1011, wherein the neural stem cells are cryopreserved.
- (Currently Amended) An isolated neuron, which is obtained prepared by the method of claim 8
- 14. (Currently Amended) The isolated neuron according to claim 13, wherein the isolated neuron cell expresses at least one kind-member selected from the group consisting of class III β tubulin, neurofilament, tyrosine hydroxylase, glutamate decarboxylase and choline acetyltransferase.
- 15. (Currently Amended) An isolated glial cell, which is obtained prepared by the method according to claim 9.
- 16. (Currently Amended) A cell pharmaceutical composition comprising, as an active ingredient, isolated neural stem cells which are <u>differentiated-produced</u> from embryonic stem cells by the method according to claim 1.
- 17. (Currently Amended) A cell pharmaceutical composition comprising, as an active ingredient, an isolated neuron obtained by the method according to claim 8 and a pharmacologically acceptable carrier.

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18. (Currently Amended) A cell pharmaceutical composition comprising, as an active

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ingredient, isolated glial cells obtained by the method according to claim 9 and a

pharmacologically acceptable carrier.

19. (Cancelled)

20. (New) A method for producing isolated neural cells, wherein the method comprises:

culturing primate embryonic stem cells for a period of 2-4 days in suspension, in the presence of

a media consisting essentially of an astrocyte conditioned medium, a medium including the supernatant of the culture of astrocytes, or a basal medium which has metabolites from an

astrocyte cell culture, to produce said isolated neural cells.

21. (New) The method of claim 1, wherein prior to said culturing in suspension, said

primate embryonic stem cells are grown on a feeder cell layer, and a colony of embryonic stem

cells is mechanically detached and added to an medium comprising astrocyte conditioned

medium to obtain culture in suspension.

22. (New) The method according to claim 1 wherein said primate embryonic stem cells

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